

# Variability in cone and seed characteristics and seed testing in various provenances of Himalayan spruce (*Picea smithiana*)

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**Abstract:** We investigated the effect of provenance variation on cone and seed morphology and germination behaviour under different pretreatments in *Picea smithiana*. Three categories of cones were recognized: large (13.18 cm × 3.30 cm–15.42 cm × 4.35 cm), medium (10.85 cm × 3.93 cm–12.18 cm × 3.98 cm) and small (7.69 cm × 3.06–10.98 cm × 3.39 cm). Significant variation was observed for various cone and seed characteristics. Seeds moisture content varied from 44.48% to 56.91%, seed size from 0.64 cm × 0.31 cm (largest seed) to 0.49 cm × 0.10 cm (smallest seed), and the seed weight from 2.45 g per 100 seeds to 1.36 g per 100 seeds on fresh weight basis of *P. smithiana*. The highest seed germination (72.0±7.53%) at 10°C was observed under chilling treatment in Tapovan provenance, while the minimum (15.0±5.71%) seed germination was recorded at 25°C under control set in Tapovan provenance. Further among all the sources Pandukeshwar consistently had the highest average (38%) percent germination across all treatments. Compared to other temperature regimes and pretreatments, seeds subjected to 10°C and chilling treatment had the highest germination and took the least time for germination irrespective of provenance effects. Altitude (provenance) had little or no relationship with germination following laboratory treatments. Seeds from Tapovan provenance subjected at 10°C following chilling treatment may be suggested for further multiplication of this species.

**Keywords:** Dormancy; Garhwal Himalaya; germination value; seed sources; seed weight

## Introduction

The Himalayan spruce (*P. smithiana* Wallich) of family coniferae is a tall evergreen conifer with a characteristic conical crown,

horizontal or drooping branches, dark green foliage and found distributed throughout the western Himalaya between 2 150 m and 3 350 m elevations. It is rarely found as pure crop over extensive areas, but more commonly mixed with *Abies pindrow*, *Cedrus deodara*, *Pinus wallichiana*, *Quercus semecarpifolia*, *Acer caesium*, *Aesculus indica*, *Prunus padus*, *Ulmus wallichiana* and other species (Troup 1921). The large scale felling of *P. smithiana* for timber, raw material and various other purposes resulted in massive destruction of this species. To reduce the imbalance of this species in various ecosystems and increase the forest cover, studies on seed characteristics and germination behaviour were considered of great importance to understand its regeneration pattern in the nature. Natural regeneration in Himalayan spruce is, however, generally deficient and conspicuous in many areas by its absence. Since then the problem of natural regeneration in the species was constantly engaging the attention of the forest scientists. Number of factors, namely lack of adequate light on the forest floor in the forests managed under selection system (Troup 1921), thick layer of humus (Troup 1921; Taylor et al. 1934; Glover 1936; Kaul 1970), accumulation of debris (Hafizullah 1970), dense weed growth (Troup 1921), and continuous grazing (Redcliffe 1906; Flewett 1930; Sufi 1970), are considered responsible for the absence of natural regeneration. Infrequent seed years and low germinative capacity of the seeds are also important factors contributing to the absence of natural regeneration in this species. Germination physiology generally indicates a temperature range and an optimum temperature at which the highest percent of germination occurs within the shortest period of time. Further, each species has its own temperature requirement for germination, suggesting a possible genetic control (Khullar et al. 1991).

Successful afforestation requires large quantities of seeds and great care, right from collection to germination. Seeds of *P. smithiana* exhibit a great range of variation in shape, size, colour and seed coat. Therefore, the knowledge of seed morphology is advantageous for artificial regeneration as it can influence the collection, storage processing and the treatment of seeds (Uniyal et al. 2000; ISTA 1999). In view of the increased reliance on artificial regeneration, seed production assumes significant im-

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portance. Infrequent seed years and low germination capacity of seeds in this species necessitates collection and testing as much quantities of seeds as possible. Therefore, the study aim was to understand the variation in morphological characteristics of cones and seeds, enhancement of seed germination by using various pre-treatments and temperature regimes in five seed sources of *P. smithiana* from Garhwal Himalaya.

## Materials and methods

The seed sources were located between 29°26' N–31°28' N and 77° 49' E–80°06' E, from 2 595 m to 2 880 m in the moist temperate and sub alpine spruce forests of Garhwal Himalaya (Table 1 and Fig. 1). To assess different sizes of cones, seed characteristics and their germinability, the mature cones were collected from wide range of habitats in Chamoli district of Garhwal Himalaya. The majority of rainfall in this region, occurs during monsoon period (i.e., from June to September), and the area

represents typical temperate climate (Table1).

Hundred mature cones from each seed source were collected during from October to November 2003. The cones were air dried and seeds were extracted manually. Five samples of ten cones drawn from each seed source in each size class ( $5 \times 10 \times 3 = 150$ ), which were used to record the cone size (length and thickness), the number of healthy and aborted seeds of per cone, and the moisture content of cone. Cone length and thickness were measured by scale (for length) and Vernier calliper (for thickness) in cm, up to two decimal places. Five samples of 100 seeds were drawn from each seed source in each size class ( $5 \times 100 \times 3 = 1500$ ), which were measured for their maximum length and width (in cm, up to two decimal places) by using Vernier calliper, to study the variation in seed size. Fresh seed weight of ten randomly taken samples of each seed source, per size class consisting of 100 seeds each ( $5 \times 100 \times 3 = 1500$ ) were obtained as per International Seed Testing Association (1999) rules using an electronic top pan balance (Model No. 7301A – Anamed).

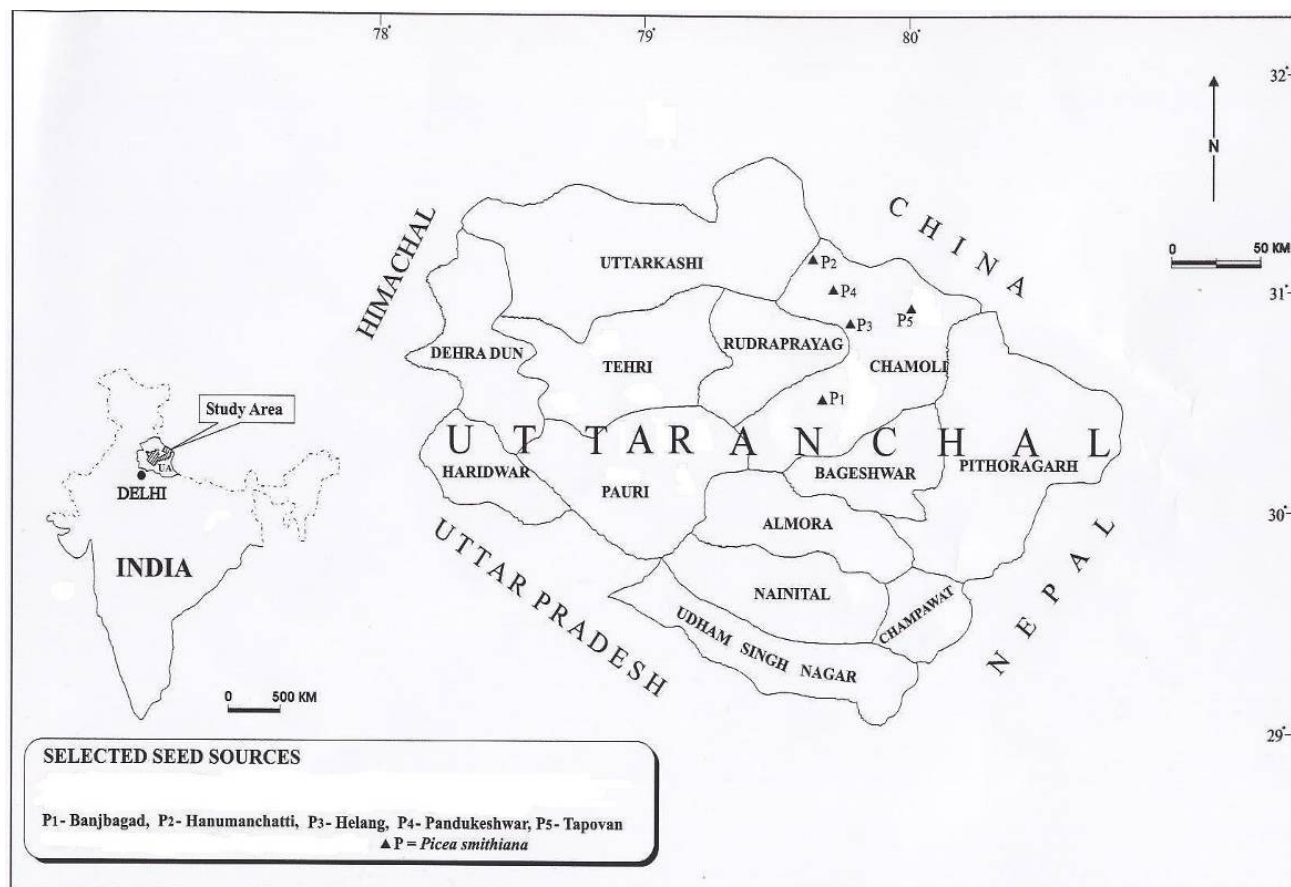


Fig. 1 Location map of the study area

### Temperature treatment

Five replicates, each of 100 randomly selected seeds, from each seed source were tested for germination. Seeds were placed on Petri dishes of 9 cm diameter containing double layered mois-

tened Whatman No. 1 filter paper. Dishes were placed in germination chambers (Model No. 8LT-SGL CALTAN) at four constant temperature regimes: 10°C, 15°C, 20°C and  $(25 \pm 2)^\circ\text{C}$  in the dark. Filter papers were kept moistened with few drops of double-distilled water.

## Pretreatments

Five replicates, each of 100 randomly selected seeds, from each seed source were used for the pretreatments as follows:

Treatment 1: Soaking seeds in distilled water at room temperature ( $25\pm 2^\circ\text{C}$ ) for 24 h (control).

Treatment 2: Soaking seeds in Gibberellic acid ( $\text{GA}_3$   $100\ \mu\text{L}\cdot\text{L}^{-1}$ ) at room temperature ( $25\pm 2^\circ\text{C}$ ) for 24 h.

Treatment 3: Soaking seeds in Hydrogen peroxide ( $\text{H}_2\text{O}_2$  1% v/v) at room temperature ( $25\pm 2^\circ\text{C}$ ) for 24 h.

Treatment 4: Seeds treated with Gibberellic acid ( $\text{GA}_3$   $100\ \mu\text{L}\cdot\text{L}^{-1}$ ) as above at room temperature ( $25\pm 2^\circ\text{C}$ ) for 24 h, then chilled for 15 days (at  $2-3^\circ\text{C}$ ). (Chilling of seeds was carried out in folded over polythene bags at  $2-3^\circ\text{C}$  for 15 days (as per method suggested by Tompsett and Pritchard (1998). The chilled seeds were then subjected to various temperature

treatments.).

Germination test duration was 28 days under each temperature regime and pretreatment. Germinated seeds (criterion: emergence of plumule and radicle) were counted daily for 28 days. Observations were made on the percent of germination. All the data were subjected to ANOVA, and further mean values were analyzed by using least significant difference (LSD). The relationship of altitude and percent germination under each treatment was calculated using Pearson's correlation coefficient ( $r$ ), and analysis of variance was computed following Sharma (1998).

Germination Value ( $GV$ ) was calculated following Czabator (1962):

$$GV = PV \times MDG \quad (1)$$

where,  $PV$  is the peak value of germination, and  $MDG$  is the mean daily germination.

**Table1. Geographical and meteorological description of the seed sources of *P. smithiana*.**

Provenance	District	Altitude (m)	Latitude	Longitude	Temperature ( $^\circ\text{C}$ )		Mean annual rainfall (mm)
					Min.	Max.	
Banjbagad	Chamoli	2775	$30^\circ 15' \text{ N}$	$79^\circ 34' \text{ E}$	1.13	30.8	1276.00
Hanumanchatti	Chamoli	2880	$30^\circ 41' \text{ N}$	$79^\circ 30' \text{ E}$	-0.10	25.3	2098.00
Helang	Chamoli	2595	$30^\circ 33' \text{ N}$	$79^\circ 37' \text{ E}$	1.34	28.9	1860.00
Pandukeshwar	Chamoli	2657	$30^\circ 31' \text{ N}$	$79^\circ 32' \text{ E}$	-0.91	27.0	1932.00
Tapovan	Chamoli	2798	$30^\circ 31' \text{ N}$	$79^\circ 36' \text{ E}$	-0.84	24.5	1892.00

## Results

### Cone and seed traits

The large, medium and small sized cone length in different seed sources varied from  $13.18\pm 0.116$  cm (Pandukeshwar) to  $15.42\pm 0.121$  cm (Helang),  $10.85 \pm 0.088$  cm (Banjbagad) to  $12.18 \pm 0.076$  cm (Tapovan), and from  $7.69\pm 0.158$  cm (Helang) to  $10.98\pm 0.077$  cm (Tapovan), respectively. Cone thicknesses in different seed sources varied from  $3.30\pm 0.023$  cm (Pandukeshwar) to  $4.43\pm 0.027$  cm (Hanumanchatti),  $2.99\pm 0.018$  cm (Pandukeshwar) to  $3.98\pm 0.034$  cm (Tapovan), and from  $2.75\pm 0.025$  cm (Hanumanchatti) to  $3.39\pm 0.029$  cm (Tapovan) for large, medium, and small size cones, respectively. The number of scales per cone in the same way ranged between  $198\pm 1.566$  (Tapovan) to  $232.72\pm 0.747$  (Pandukeshwar),  $158.72\pm 3.42$  (Helang) to  $191.48\pm 1.29$  (Banjbagad), and  $135.38\pm 1.02$  (Tapovan) to  $173.9\pm 1.67$  (Banjbagad) for large, medium and small sized cones, respectively. Similarly, the number of seeds of per cone for large, medium and small size cones in different seed sources were recorded as  $252.86\pm 7.15$  (Tapovan) to  $428.46\pm 2.48$  (Pandukeshwar),  $240.09\pm 5.03$  (Tapovan) to  $370.92\pm 2.99$  (Hanumanchatti), and  $163.68\pm 4.46$  (Tapovan) to  $315.72 \pm 4.09$  (Banjbagad), respectively. The cone moisture content in large, medium and small sized cones varied from  $44.48\pm 1.35\%$  (Helang) to  $56.91\pm 1.28\%$  (Pandukeshwar),  $45.071.42\%$  (Pandukeshwar) to

$52.97\pm 0.79\%$  (Banjbagad), and  $51.13\pm 1.26\%$  (Hanumanchatti) to  $53.32\pm 1.67\%$  (Tapovan), respectively (Table 2).

The maximum lengths for sized seeds, large ( $0.64\pm 0.007$  cm), medium ( $0.50\pm 0.006$  cm), and small ( $0.41\pm 0.003$  cm), were recorded in Pandukeshwar seed source, while the minimum lengths for sized seeds, large ( $0.500\pm 0.003$  cm), medium ( $0.40\pm 0.002$  cm), and small ( $0.29\pm 0.004$  cm), were recorded in Hanumanchatti seed source. Similarly, the maximum seed widths for sized seeds, large ( $0.32\pm 0.006$  cm), medium ( $0.31\pm 0.004$  cm), and small ( $0.27\pm 0.007$  cm), were observed in Tapovan seed sources, and minimum seed widths for sized seeds, large ( $0.28\pm 0.005$  cm), medium ( $0.27\pm 0.005$  cm), and small ( $0.20\pm 0.006$  cm) was recorded in Helang seed source. The seed thicknesses among different seed sources for large, medium and small sized seeds were  $0.17\pm 0.005$  cm (Hanumanchatti) to  $0.21\pm 0.004$  cm (Tapovan),  $0.17\pm 0.005$  cm (Banjbagad) to  $0.20\pm 0.003$  cm (Tapovan), and  $0.10\pm 0.002$  cm (Helang) to  $0.19\pm 0.002$  cm (Banjbagad), respectively. The weight of 100 seeds with wings for large, medium and small sized seeds varied from  $1.49\pm 0.003$  g (Banjbagad) to  $2.45\pm 0.004$  g (Hanumanchatti),  $1.18\pm 0.002$  g (Pandukeshwar) to  $2.29\pm 0.002$  g (Hanumanchatti), and from  $0.85\pm 0.003$  g (Pandukeshwar) to  $1.36\pm 0.003$  g (Hanumanchatti), respectively, in various selected seed sources of *P. smithiana*, whereas, the weight of 100 seeds without wings for large, medium and small sized seeds ranged between  $1.234\pm 0.003$  g (Banjbagad) to  $2.05\pm 0.003$  g (Hanumanchatti),  $0.95\pm 0.002$  g (Pandukeshwar) to  $1.96\pm 0.003$  g (Hanumanchatti), and  $0.55\pm 0.003$  g (Pandukeshwar) to  $0.98\pm 0.003$  g (Hanumanchatti), respectively (Ta-

ble 2).

**Table 2.** Cone and seed characteristics of different provenances of *P. smithiana*

Provenance	Length (cm)			Thickness (cm)			No. of Scales/cone			No. of Seeds/cone			Moisture (%)		
	Large>13cm	Medium 10-12cm	Small<10cm	Large	Medium	Small	Large	Medium	Small	Large	Medium	Small	Large	Medium	small
<b>Cone characteristics</b>															
Banjbagad	13.18 <sup>b</sup> ±0.11	10.85 <sup>b</sup> ±0.09	9.01 <sup>ab</sup> ±0.07	3.93 <sup>ab</sup> ±0.03	3.53 <sup>ab</sup> ±0.01	3.05 <sup>ab</sup> ±0.02	214.24 <sup>b</sup> ±0.74	191.48 <sup>a</sup> ±1.29	173.9 <sup>a</sup> ±1.67	412.38 <sup>a</sup> ±2.07	362.3 <sup>a</sup> ±5.00	315.72 <sup>a</sup> ±4.09	52.45 <sup>ab</sup> ±1.76	52.97 <sup>a</sup> ±0.79	52.28 <sup>ab</sup> ±0.93
Hanu-man-Chatti	14.19 <sup>ab</sup> ±0.07	10.88 <sup>b</sup> ±0.08	7.77 <sup>b</sup> ±0.12	4.43 <sup>a</sup> ±0.03	3.26 <sup>ab</sup> ±0.04	2.75 <sup>b</sup> ±0.03	206.08 <sup>b</sup> ±1.25	189.4 <sup>a</sup> ±0.67	149.4 <sup>ab</sup> ±3.07	389.24 <sup>ab</sup> ±4.05	370.92 <sup>a</sup> ±2.99	264.86 <sup>ab</sup> ±4.16	50.23 <sup>ab</sup> ±1.77	51.01 <sup>a</sup> ±1.38	51.13 <sup>b</sup> ±1.26
Helang	15.42 <sup>a</sup> ±0.12	11.59 <sup>ab</sup> ±0.10	7.69 <sup>b</sup> ±0.16	4.35 <sup>a</sup> ±0.03	3.80 <sup>a</sup> ±0.03	3.06 <sup>ab</sup> ±0.04	218.76 <sup>ab</sup> ±1.50	158.72 <sup>b</sup> ±3.42	162.96 <sup>ab</sup> ±2.12	418.88 <sup>a</sup> ±2.82	287.28 <sup>ab</sup> ±3.12	280.5 <sup>ab</sup> ±4.27	44.48 <sup>b</sup> ±1.35	49.53 <sup>ab</sup> ±1.76	52.63 <sup>ab</sup> ±1.10
Pandukeshwar	13.18 <sup>b</sup> ±0.12	11.06 <sup>b</sup> ±0.07	9.45 <sup>ab</sup> ±0.10	3.30 <sup>b</sup> ±0.02	2.99 <sup>b</sup> ±0.02	2.78 <sup>b</sup> ±0.02	232.72 <sup>a</sup> ±0.75	186.7 <sup>a</sup> ±1.68	140.12 <sup>b</sup> ±1.76	428.46 <sup>a</sup> ±2.48	355.34 <sup>a</sup> ±3.37	256.92 <sup>ab</sup> ±3.41	56.91 <sup>a</sup> ±1.28	45.07 <sup>b</sup> ±1.42	52.60 <sup>ab</sup> ±1.51
Tapovan	14.94 <sup>ab</sup> ±0.13	12.18 <sup>a</sup> ±0.08	10.98 <sup>a</sup> ±0.08	4.21 <sup>a</sup> ±0.02	3.98 <sup>a</sup> ±0.03	3.39 <sup>a</sup> ±0.03	198.00 <sup>b</sup> ±1.57	171.92 <sup>ab</sup> ±0.89	135.38 <sup>b</sup> ±1.02	252.86 <sup>b</sup> ±7.15	240.09 <sup>b</sup> ±5.03	163.68 <sup>b</sup> ±4.46	46.19 <sup>b</sup> ±1.12	47.09 <sup>ab</sup> ±1.82	53.32 <sup>a</sup> ±1.67
'r' Vs Altitude	-0.183	-0.176	0.155	0.369	-0.033	-0.009	-0.712	0.592	-0.160	-0.440	0.221	0.241	0.054	0.391	-0.488
<b>Seed characteristics</b>															
Provenance	Seeds length (cm)			Seed width (cm)			Seed thickness (cm)			100 seed weight with wings (g)			100 seed weight without wings (g)		
	Large>4cm	Medium 4-5cm	Small<4cm	Large	Medium	small	Large	Medium	Small	Large	Medium	Small	Large	Medium	Small
Banjbagad	0.51 <sup>b</sup> ±0.004	0.40 <sup>b</sup> ±0.003	0.36 <sup>ab</sup> ±0.006	0.31 <sup>ab</sup> ±0.004	0.27 <sup>b</sup> ±0.005	0.21 <sup>ab</sup> ±0.005	0.20 <sup>a</sup> ±0.002	0.17 <sup>b</sup> ±0.005	0.19 <sup>a</sup> ±0.002	1.49 <sup>b</sup> ±0.003	1.27 <sup>b</sup> ±0.003	0.87 <sup>b</sup> ±0.12	1.23 <sup>b</sup> ±0.003	1.04 <sup>b</sup> ±0.001	0.64 <sup>ab</sup> ±0.004
Hanu-man-Chatti	0.50 <sup>b</sup> ±0.003	0.40 <sup>b</sup> ±0.002	0.29 <sup>b</sup> ±0.004	0.29 <sup>bc</sup> ±0.003	0.28 <sup>b</sup> ±0.004	0.20 <sup>b</sup> ±0.002	0.17 <sup>b</sup> ±0.005	0.17 <sup>b</sup> ±0.006	0.10 <sup>b</sup> ±0.002	2.45 <sup>a</sup> ±0.004	2.29 <sup>a</sup> ±0.002	1.36 <sup>a</sup> ±0.003	2.05 <sup>a</sup> ±0.003	1.96 <sup>a</sup> ±0.003	0.98 <sup>a</sup> ±0.003
Helang	0.59 <sup>ab</sup> ±0.002	0.48 <sup>ab</sup> ±0.005	0.40 <sup>a</sup> ±0.004	0.28 <sup>c</sup> ±0.005	0.27 <sup>b</sup> ±0.005	0.20 <sup>b</sup> ±0.006	0.20 <sup>a</sup> ±0.002	0.20 <sup>a</sup> ±0.028	0.10 <sup>b</sup> ±0.002	2.04 <sup>ab</sup> ±0.003	1.87 <sup>a</sup> ±0.003	1.32 <sup>b</sup> ±0.002	1.64 <sup>ab</sup> ±0.006	1.43 <sup>ab</sup> ±0.002	0.98 <sup>a</sup> ±0.138
Pan-dukeshwar	0.64 <sup>a</sup> ±0.007	0.50 <sup>a</sup> ±0.006	0.41 <sup>a</sup> ±0.003	0.31 <sup>ab</sup> ±0.005	0.29 <sup>ab</sup> ±0.003	0.26 <sup>ab</sup> ±0.006	0.20 <sup>a</sup> ±0.003	0.20 <sup>a</sup> ±0.002	0.14 <sup>ab</sup> ±0.007	1.55 <sup>ab</sup> ±0.002	1.18 <sup>b</sup> ±0.002	0.85 <sup>b</sup> ±0.003	1.34 <sup>b</sup> ±0.004	0.95 <sup>b</sup> ±0.002	0.55 <sup>b</sup> ±0.003
Tapovan	0.60 <sup>ab</sup> ±0.005	0.50 <sup>a</sup> ±0.002	0.40 <sup>a</sup> ±0.002	0.32 <sup>a</sup> ±0.006	0.31 <sup>a</sup> ±0.004	0.27 <sup>a</sup> ±0.007	0.21 <sup>a</sup> ±0.004	0.20 <sup>a</sup> ±0.003	0.14 <sup>ab</sup> ±0.007	2.27 <sup>ab</sup> ±0.002	1.88 <sup>a</sup> ±0.011	0.97 <sup>b</sup> ±0.005	1.94 <sup>ab</sup> ±0.003	1.55 <sup>ab</sup> ±0.011	0.76 <sup>ab</sup> ±0.004
'r' Vs Altitude	-0.682	-0.608	-0.771**	0.289	0.261	-0.045	-0.520	-0.692	0.117	0.156	0.455	0.090	0.520	0.577	0.135

After “±” is SE

The seed germination in different provenances after pre-soaking treatments under different temperature regimes, (i.e., 10°C, 15°C, 20°C and 25°C) yielded significant differences. The analysis of variance revealed significant differences amongst different seed sources (Table 3). In *P. smithiana*, the maximum germination of seeds was observed at 10°C (72.0±7.53%) followed by 15°C (55.0±4.19%) in Tapovan seed source under chilling treatment, whereas, the minimum germination of seeds (17±1.22) was found at 20°C in Pandukeshwar under H<sub>2</sub>O<sub>2</sub> (1% v/v) treatment, followed by (15±5.71) at 25°C in Tapovan source under GA<sub>3</sub> (100 µL·L<sup>-1</sup>) treatment. On the other hand, the maximum germination values at 10°C, 15°C, 20°C and 25°C were observed in Tapovan (1.26±0.31 at 10°C, and 0.84±0.13 at 15°C) and Pandukeshwar (1.57±0.53 at 20°C and 1.08±0.34 at 25°C) seed sources Table 4).

Statistical analysis reveals that all most all the cone and seed traits among the provenances were found statistically significant (Table 3 and Table 5).

## Discussion

As this species experienced colder climate and aspects during its whole life span, thus seed germination pattern also reveals the hierarchy to be flourished under lower temperatures after chilling treatment. Various study results suggest that pretreatments are species specific (Khullar et al. 1991), generally hot water treatment is reported for *Grewia oppositifolia* (Uniyal et al. 2000), *Dalbergia sissoo* (Uniyal et al. 2007), *Albizia chinensis* (Dhanai et al. 2009). The correlation coefficients between different cone and seed traits among various provenances of *P. smithiana* reveals that the cone length has a strong correlation with cone diameter ( $r=0.7595$ ) and 100 seeds weight ( $r=0.6157$ ), while a negative correlation with seed width ( $r=-0.4421$ ). The cone diameter shows strong positive correlation with 100 seeds weight ( $r=0.7295$ ) and negative correlation with seeds/cone ( $r=-0.3409$ ) and seed thickness ( $r=-0.3575$ ). The seed length has positive correlation with seed width ( $r=0.2732$ ), seed thickness

( $r = 0.6803$ ) and cone moisture percent ( $r = 0.0750$ ). The cone width on the other hand shows a significant correlation with seed thickness ( $r = 0.5417$ ), and cone moisture content percent ( $r = 0.3827$ ). On the other hand, the seed thickness exhibited negative correlation with cone moisture percent ( $r = -0.1458$ ) (Table 6). Further, the percent germination and germination value were less or no influenced under any specific treatment when correlated with altitude of the provenances. Significant correlations (with altitude) were observed for germination under control set (negatively significant) at 10 and 15°C, significantly positive correlation for germination was recorded for GA<sub>3</sub>, H<sub>2</sub>O<sub>2</sub> and chilling treatments (under 25°C), while germination value was positively correlated with altitude of provenances under GA<sub>3</sub> (under 15°C and 20°C), H<sub>2</sub>O<sub>2</sub> (under 25°C) and chilling treatment (under 15°C) (Table 4). Correlation studies clearly suggested that altitude does not play significant role by the treatment for enhancing the percent germination. However, it may be influenced by seed size and weight.

**Table3. Analysis of variance for cone and seed characteristics**

Source of variation	df	SS	MSS	F-ratio	F-critical		CD	
					5%	1%	5%	1%
Cone characteristics. Main plot								
Provenances	4	4.15	1.1	6.87*	3.84	7.01	0.58	0.85
Cone size	2	6.48	3.24	20.25**	4.48	8.85	0.75	1.1
Error[a]	8	1.24	0.16					
Split plot								
Cone characteristic	3	2.96	0.98	5.76**	2.89	4.42	0.49	0.65
Cone size X Cone characteristic	6	1.79	0.29	1.76	2.37	3.38	0.37	0.61
Split plot Error[b]	36	4.25	0.12					
Total	59	20.87						
Seed characteristics (with wings)								
Seed weight	2	1.52	0.758	28.004**	4.48	8.649	0.31	0.45
Provenances	4	2.11	0.527	19.48**	3.84	7.006	0.24	0.35
Error	8	0.22	0.027					
Total	14	3.84						
Seed characteristics (without wings)								
Seed weight	2	1.25	0.622	15.19**	4.48	8.649	0.38	0.55
Provenances	4	1.59	0.399	9.74**	3.84	7.006	0.29	0.43
Error	8	0.32	0.041					
Total	14	3.17						

\*Significant at 5% and \*\*significant at 1%

The extent of variation in the seed sources for all cone parameters were large, and the factors responsible for these differences may be edaphic and/or climatic (especially fertility and moisture content) prevalent at the sites of parent seed sources. The differences among seed sources may be attributed to variation due to natural selection for different traits in their natural habitats. Some workers advocated that, grading of seeds, better storage conditions and evaluation of germination potential prior to sowing may be an advantage (Rawat et al. 2006; Rawat et al. 2006). Grading of the seeds also proved its utility for raising quality seedlings of horticultural crops (Srimathi et al. 2001).

The cones, seeds and germination traits may be largely under maternal influences and are also strongly controlled by age, general health, and micro and macro habitats of the parent trees Isik (1986). Similar results with respect to cone/pod/fruit and seed parameters were reported in *Pinus roxburghii* by (Todaria and Uniyal 2003; Ghildiyal and Sharma 2005), *Grewia oppositifolia* (Uniyal et al. 2001 & 2003), *Albizia chinensis* (Dhanai et al. 2004), *Abies pindrow* (Rawat et al. 2008) and *Cupressus torulosa* (Rawat et al. 2008). These workers concluded that cone/pod/seed characteristics of various species show highly significant differences between geographical locations, but were not significantly correlated with latitude or longitude of the seed sources. Among the seed sources of *P. smithiana*, seed weight displayed wide variation (Table 2), which may control the germination and seeding growth, and usually heavier seeds germinate faster than the lighter ones, as was also reported for *Grewia oppositifolia* (Uniyal et al. 2003), *Albizia chinensis* (Dhanai et al. 2004; Isik 1986), *P. smithiana* (Singh et al. 1990) and *Abies pindrow* (Singh and Shah 1992). Therefore, seed weight for understanding the geographical variations was advocated by many scientists, due to lowest plasticity in this trait (Harper et al. 1970). Uniyal et al. (2002), Dhanai et al. (2003), Todaria and Uniyal (2003) and Singh et al. (2006) considered the seed size to be one of the least plastic traits, and they showed that in some cases seed size could be used to identify seed collection lots of unknown provenances. Higher seed weight leads to lesser number of empty seeds and good nutrition condition for growth and development of healthy seedlings. Uniyal et al. (2003) and Dhanai et al. (2004) found positive correlation between germination vigour and seedling dry weight with seed weight. Variation in seed weight directly influence the germination capacity, as heavy seeds germinate early and achieve greater germination percentage than small and medium sized seeds in *Quercus semiserrata* (Khan and Uma Shankar 2001). This may be due to greater stocks of food and energy in heavy seeds than small and intermediate weight seeds, which provide comparatively lesser readily available energy to stimulate germination (Uniyal et al. 2003; Dhanai et al. 2004). Many workers have emphasised that the edaphic and climatic factors of the places of origin are the most crucial factors affecting seed traits. Apart from these factors, age, vigour, crown exposure and the genotype of the tree are also important in explaining seed source dependent variations (Uniyal et al. 2002 and 2003; Dhanai et al. 2003 and 2004). However, in a situation like the present study, where seeds of different localities were collected from trees of approximately the same age and crown exposure, the differences in seed parameters may be attributed to the different genetic architectures, developed as a result of adaptation in diverse environmental conditions and was found existing throughout their distributional range (Salazar 1989). These differences for cone and seed traits (Table 2) indicate the need for the recombination breeding in future for combining the superior traits in a single genotype.

In most plant systems, the rate of germination is strongly temperature dependent (Khullar et al. 1991). The International Seed Testing Association (ISTA, 1999) recommended temperatures between 20°C and 30°C for optimum seed germination in most

of the forest tree species (Saklani et al. 2001); *Quercus floribunda* obtained optimum seed germination at 25°C. Gupta and Pattanath (1976) framed germination test rules for 62 tropical and temperate seeds and suggested that some species like *Abies*

*pindrow*, *Cupressus arizonica*, *Fraxinus micrantha* and *P. smithiana* are temperature-sensitive and require alternating day and night temperatures of 30°C and 20°C for optimum germination.

**Table 4.** Effect of different provenance, pretreatments and temperature regimes on seed germination and germination value of *P. smithiana*

Provenances	Control		GA <sub>3</sub> (100 µL·L <sup>-1</sup> )		H <sub>2</sub> O <sub>2</sub> (1%v/v)		Chilling	
	G (%)	GV	G (%)	GV	G (%)	GV	G (%)	GV
<b>10°C</b>								
Banjbagad	31±1.86 <sup>ab</sup>	0.54±0.11 <sup>b</sup>	42±5.62 <sup>b</sup>	0.83±0.29 <sup>b</sup>	32±3.75 <sup>ab</sup>	0.48±0.09 <sup>b</sup>	52±3.40 <sup>b</sup>	0.73±0.18 <sup>b</sup>
Hanumanchatti	31±6.61 <sup>ab</sup>	0.83±0.29 <sup>ab</sup>	53±4.07 <sup>ab</sup>	0.93±0.18 <sup>b</sup>	32±9.72 <sup>ab</sup>	0.91±0.26 <sup>a</sup>	63±6.0 <sup>ab</sup>	1.17±0.2 <sup>ab</sup>
Helang	37±2.25 <sup>a</sup>	0.69±0.09 <sup>b</sup>	45±5.01 <sup>ab</sup>	1.05±0.2 <sup>ab</sup>	33±3.0 <sup>a</sup>	0.47±0.06 <sup>b</sup>	54±8.59 <sup>b</sup>	0.82±0.27 <sup>b</sup>
Pandukeshwar	36±3.68 <sup>a</sup>	1.26±0.30 <sup>a</sup>	52±3.75 <sup>ab</sup>	1.07±0.2 <sup>ab</sup>	31±4.01 <sup>b</sup>	0.54±0.11 <sup>b</sup>	59±5.3 <sup>ab</sup>	1.48±0.2 <sup>a</sup>
Tapovan	27±3.75 <sup>b</sup>	0.68±0.09 <sup>b</sup>	56±6.01 <sup>a</sup>	1.29±0.0 <sup>a</sup>	32±5.84 <sup>ab</sup>	0.50±0.19 <sup>b</sup>	72±7.5 <sup>a</sup>	1.26±0.3 <sup>ab</sup>
'r' vs Altitude	-0.814*	-0.270	-0.429	0.178	-0.423	-0.292	-0.222	0.270
<b>15°C</b>								
Banjbagad	29±2.92 <sup>ab</sup>	0.49±0.09 <sup>b</sup>	38±3.00 <sup>a</sup>	0.07±0.22 <sup>b</sup>	27±4.37 <sup>ab</sup>	0.37±0.10 <sup>b</sup>	32±5.16 <sup>b</sup>	0.31±0.04 <sup>b</sup>
Hanumanchatti	22±2.55 <sup>b</sup>	0.41±0.10 <sup>b</sup>	30±4.48 <sup>b</sup>	0.64±0.13 <sup>a</sup>	27±3.00 <sup>ab</sup>	0.42±0.12 <sup>ab</sup>	41±5.80 <sup>ab</sup>	0.50±0.09 <sup>ab</sup>
Helang	29±1.12 <sup>ab</sup>	0.59±0.05 <sup>ab</sup>	38±6.65 <sup>a</sup>	0.59±0.12 <sup>ab</sup>	31±5.35 <sup>ab</sup>	0.58±0.22 <sup>a</sup>	35±5.25 <sup>b</sup>	0.38±0.10 <sup>b</sup>
Pandukeshwar	30±1.58 <sup>a</sup>	0.61±0.05 <sup>ab</sup>	38±2.00 <sup>a</sup>	0.78±0.13 <sup>a</sup>	34±2.92 <sup>a</sup>	0.54±0.11 <sup>ab</sup>	50±3.17 <sup>ab</sup>	0.99±0.32 <sup>a</sup>
Tapovan	22±2.00 <sup>b</sup>	0.82±0.1 <sup>a</sup>	36±5.58 <sup>ab</sup>	0.61±0.05 <sup>a</sup>	24±4.31 <sup>b</sup>	0.37±0.10 <sup>b</sup>	55±4.19 <sup>a</sup>	0.84±0.13 <sup>ab</sup>
'r' vs Altitude	-0.790**	-0.241	0.387	0.736**	-0.227	0.063	-0.543	-0.066
<b>20°C</b>								
Banjbagad	29±6.22 <sup>ab</sup>	0.57±0.13 <sup>ab</sup>	36±3.68 <sup>b</sup>	0.85±0.18 <sup>b</sup>	35±2.2 <sup>ab</sup>	0.87±0.15 <sup>ab</sup>	39±4.31 <sup>b</sup>	0.49±0.07 <sup>b</sup>
Hanumanchatti	26±4.31 <sup>ab</sup>	0.55±0.09 <sup>ab</sup>	43±3.75 <sup>ab</sup>	1.02±0.13 <sup>b</sup>	46±2.4 <sup>a</sup>	1.35±0.20 <sup>a</sup>	48±6.05 <sup>a</sup>	0.91±0.18 <sup>a</sup>
Helang	26±4.31 <sup>ab</sup>	0.46±0.08 <sup>b</sup>	43±3.75 <sup>ab</sup>	1.16±0.39 <sup>ab</sup>	46±2.45 <sup>a</sup>	0.81±0.10 <sup>ab</sup>	48±6.05 <sup>a</sup>	0.62±0.16 <sup>ab</sup>
Pandukeshwar	25±1.58 <sup>b</sup>	0.42±0.06 <sup>b</sup>	54±11.90 <sup>a</sup>	1.57±0.53 <sup>a</sup>	17±1.22 <sup>b</sup>	0.20±0.05 <sup>b</sup>	45±4.75 <sup>ab</sup>	0.41±0.15 <sup>b</sup>
Tapovan	30±2.74 <sup>a</sup>	0.71±0.21 <sup>a</sup>	42±2.00 <sup>ab</sup>	1.22±0.13 <sup>ab</sup>	23±2.55 <sup>ab</sup>	0.42±0.05 <sup>b</sup>	45±5.25 <sup>ab</sup>	0.57±0.11 <sup>ab</sup>
'r' vs Altitude	0.418	0.659	0.514	0.869*	-0.431	-0.620	0.289	0.851*
<b>25°C</b>								
Banjbagad	26±4.01 <sup>a</sup>	0.42±0.10 <sup>a</sup>	30±2.74 <sup>ab</sup>	0.41±0.07 <sup>b</sup>	25±2.74 <sup>ab</sup>	0.38±0.05 <sup>b</sup>	32±3.00 <sup>b</sup>	0.30±0.03 <sup>b</sup>
Hanumanchatti	21±1.87 <sup>ab</sup>	0.35±0.15 <sup>ab</sup>	28±4.37 <sup>ab</sup>	0.70±0.24 <sup>ab</sup>	24±2.45 <sup>ab</sup>	0.84±0.12 <sup>a</sup>	42±5.84 <sup>ab</sup>	0.49±0.12 <sup>b</sup>
Helang	21±1.87 <sup>ab</sup>	0.30±0.09 <sup>ab</sup>	28±4.37 <sup>ab</sup>	0.45±0.12 <sup>b</sup>	24±2.45 <sup>ab</sup>	0.27±0.04 <sup>b</sup>	42±5.84 <sup>ab</sup>	0.48±0.10 <sup>b</sup>
Pandukeshwar	25±3.54 <sup>a</sup>	0.41±0.15 <sup>a</sup>	35±4.48 <sup>a</sup>	0.84±0.23 <sup>a</sup>	27±10.2 <sup>a</sup>	0.55±0.30 <sup>ab</sup>	57±5.16 <sup>a</sup>	1.08±0.34 <sup>a</sup>
Tapovan	15±5.71 <sup>b</sup>	0.22±0.10 <sup>b</sup>	24±2.45 <sup>b</sup>	0.46±0.11 <sup>b</sup>	22±2.00 <sup>b</sup>	0.32±0.05 <sup>b</sup>	31±3.32 <sup>b</sup>	0.36±0.06 <sup>b</sup>
'r' vs Altitude	-0.257	-0.073	0.850*	0.407	0.998*	0.715	0.780**	0.296

After “±” is SE

**Table 5.** Analysis of variance for germination percent

Source of variation	df	SS	MSS	F-ratio	F-critical		CD	
					5%	1%	5%	1%
(c)P. smithiana Main plot								
Provenances	4	1.46	0.365	6.08**	3.26	5.41	0.34	0.47
Temperature	3	3.14	1.04667	17.43**	3.49	5.95	0.37	0.53
Error[a]	12	0.72	0.06					
Split plot								
Treatment	3	1.46	0.48667	1.87	2.79	4.26	0.72	0.95
Treatment X Temperature	9	7.45	0.82778	3.19**	2.08	2.81	0.46	0.61
Split plot Error[b]	48	12.45	0.25938					

\*Significant at 5% and \*\*significant at 1%

In the present study, the pretreatments considerably improved

germination. It was demonstrated previously that seed pretreatments were species-specific and no type of treatments was reported to be universally effective (Uniyal and Nautiyal 1995; Uniyal et al. 2000). This also indicates that seeds of a single species differ in germination especially when collected from different locations, and this phenomenon may be associated with altitude (Vera 1997).

The practical implication of this study is that seeds of this potential species should be subjected to chilling treatment for 15 days and then allowed for germination at 10°C to optimize germination. However, GA<sub>3</sub> (100 µL·L<sup>-1</sup>) treatment may also considerably enhance the percent germination of *P. smithiana* seeds. chilling treatment and GA<sub>3</sub> (100 µL·L<sup>-1</sup>) are easy to use, can treat seeds in large quantities and provides a fast, homogeneous method for obtaining the highest germination in seed without any danger to the user, lots of similar findings for *P. smithiana* were earlier reported by Rawat et al. (2006) and Rawat et al. (2006).

In this species it seems that seed source is as important as pre-treatment. Hence, seed collection should be conducted at an ap-

propriate locality, because seed quality has a definite relation with geographical variables of the seed source.

**Table 6. Correlation (*r*) between average cone and seed traits in *P. smithiana* provenances**

Variables	Cone length (cm)	Cone diameter (cm)	Scales/ Cone	Seeds/ cones	100Seed wt. (g)	Seed length (cm)	Seed width (cm)	Seed thickness (cm)
Cone length (cm)	-							
Cone dia. (cm)	0.75947**	-						
Scales/cone	-0.49576	-0.756912	-					
Seeds/cone	-0.436012	-0.340876	0.799889**	-				
100seed wt. (g)	0.615652**	0.729476**	-0.732961	-0.596445	-			
Seed length (cm)	0.104334*	-0.559853	0.46233**	-0.1259	-0.228292	-		
Seed width (cm)	-0.442071	-0.506971	-0.168899	-0.545734	-0.217205	0.273219*	-	
Seed thickness (cm)	0.13865*	-0.357543	0.096262*	-0.349672	-0.428624	0.680338**	0.541736**	-
Cone moisture (%)	-0.967873	-0.841827	0.622816**	0.474476**	-0.577022	0.075001*	0.38265*	-0.145827

\*Significant at 5% and \*\*significant at 1%

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